

AWARD NUMBER: W81XWH-16-1-0429

TITLE: Androgen Deprivation Therapy and Cognitive Impairment

PRINCIPAL INVESTIGATOR: Robert N. Pechnick, Ph.D.

CONTRACTING ORGANIZATION: Western University of Health Sciences
Pomona, CA 91766

REPORT DATE: August 2017

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

REPORT DOCUMENTATION PAGE				Form Approved OMB No. 0704-0188	
Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing this collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Department of Defense, Washington Headquarters Services, Directorate for Information Operations and Reports (0704-0188), 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302. Respondents should be aware that notwithstanding any other provision of law, no person shall be subject to any penalty for failing to comply with a collection of information if it does not display a currently valid OMB control number. PLEASE DO NOT RETURN YOUR FORM TO THE ABOVE ADDRESS.					
1. REPORT DATE August 2017		2. REPORT TYPE Annual		3. DATES COVERED 1 Aug 2016 - 31 Jul 2017	
4. TITLE AND SUBTITLE Androgen Deprivation Therapy and Cognitive Impairment				5a. CONTRACT NUMBER	
				5b. GRANT NUMBER W81XWH-16-1-0429	
				5c. PROGRAM ELEMENT NUMBER	
6. AUTHOR(S) Robert N. Pechnick, Ph.D. E-Mail: rpechnick@westernu.edu				5d. PROJECT NUMBER	
				5e. TASK NUMBER	
				5f. WORK UNIT NUMBER	
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) Western University of Health Sciences 309 East Second Street Pomona, CA 91766-1854				8. PERFORMING ORGANIZATION REPORT NUMBER	
9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES) U.S. Army Medical Research and Materiel Command Fort Detrick, Maryland 21702-5012				10. SPONSOR/MONITOR'S ACRONYM(S)	
				11. SPONSOR/MONITOR'S REPORT NUMBER(S)	
12. DISTRIBUTION / AVAILABILITY STATEMENT Approved for Public Release; Distribution Unlimited					
13. SUPPLEMENTARY NOTES					
14. ABSTRACT Androgen deprivation therapy is a well-established treatment for prostate cancer, but an important side effect of androgen deprivation therapy is impairment of memory and learning. In the hippocampus, a brain region that plays a major role in memory and learning, new nerve cells (i.e., neurons) continue to develop throughout adulthood, a process is called neurogenesis. The goal of this project is to test the hypothesize that impaired hippocampal neurogenesis underlies the androgen deprivation therapy-induced impairment of memory and learning. During the first year of the project we carried out surgeries (castration and sham castration) and implanted placebo pellets and pellets containing leuprolide (a gonadotropin-releasing hormone analog that reduces plasma testosterone levels) and flutamide (an androgen receptor antagonist). Some mice were administered bromodeoxyuridine to determine the effects of the treatments on the survival of the new nerve cells. Five weeks after surgery mice were sacrificed for immunohistochemical studies assessing marking of neurogenesis and Western blots for determining the expression of proteins associated with neurogenesis. All mice in all experimental groups have been treated and sacrificed. We are in the process of completing the processing of the tissue and the data analysis.					
15. SUBJECT TERMS Neurogenesis, neuron, hippocampus, memory, learning, testosterone, androgen, androgen deprivation, castration, prostate cancer, flutamide, leuprolide, proliferation, survival, immunohistochemistry, Western blot.					
16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRMC
Unclassified	Unclassified	Unclassified	Unclassified	7	19b. TELEPHONE NUMBER (include area code)

Table of Contents

	<u>Page</u>
1. Introduction.....	4
2. Keywords.....	4
3. Accomplishments.....	4
4. Impact.....	5
5. Changes/Problems.....	5
6. Products.....	6
7. Participants & Other Collaborating Organizations	6
8. Special Reporting Requirements.....	7
9. Appendices.....	N/A

1. INTRODUCTION:

Androgen deprivation therapy is a well-established treatment for prostate cancer, but an important side effect of androgen deprivation therapy is impairment of memory and learning, i.e., cognitive function. In the hippocampus, a brain region that plays a major role in memory and learning, new nerve cells continue to develop throughout adulthood, a process is called neurogenesis. The goal of this project is use an animal model to test the hypothesize that impaired hippocampal neurogenesis underlies the androgen deprivation therapy-induced impairment of cognitive function. There are four specific aims. Specific Aim 1 tests the hypothesis that androgen deprivation decreases hippocampal neurogenesis. Specific Aim 2 tests the hypothesis that androgen deprivation disrupts cognitive behavior. Specific Aim 3 tests the hypothesis that drugs that increase hippocampal neurogenesis reduces the effects of androgen deprivation on hippocampal neurogenesis and Specific Aim 4 tests the hypothesis that drugs that increase hippocampal neurogenesis reduces the effects of androgen deprivation on cognitive behavior. The results of the proposed might lead to the development of new strategies to optimize the physical and mental health of men with prostate cancer and improve the quality of life and well-being of prostate cancer patients and their families.

2. KEYWORDS:

Neurogenesis, neuron, hippocampus, memory, learning, testosterone, androgen, androgen deprivation, castration, prostate cancer, flutamide, leuprolide, proliferation, survival, immunohistochemistry, Western blot.

3. ACCOMPLISHMENTS:

What were the major goals of the project?

Year 1 Goals As Listed In Approved Statement of Work (SOW)

Specific Aim 1: To test the hypothesis that androgen deprivation decreases hippocampal neurogenesis.	
Major Task 1: Treat Animals	Months
Subtask 1: Submit documents for ACURO approvals	1-4
Milestone(s) Achieved: Obtain ACURO approval	4 (Complete)
Subtask 2: Carry out surgeries and pellet implantation [4 treatment groups (sham/placebo pellet, castration/placebo pellet, sham/flutamide pellet and sham/leuprolide pellet) x 2 subgroups (IHC and Western blot studies) x 8 mice/group = 64 mice]	4-5
Milestone(s) Achieved: Surgeries completed	5 (Complete)
Major Task 2: Sacrifice Animals, Tissue Processing and Data Analysis	
Subtask 1: Sacrifice Animals	7
Milestone(s) Achieved: Animals sacrificed	7 (Complete)
Subtask 2: Process tissue	7-11
Milestone(s) Achieved: Tissue processing completed	11 (80% Complete)
Subtask 3: Data analysis	11-12
Milestone(s) Achieved: Data analysis completed	12 (80% Complete)

What was accomplished under these goals?

The first year was devoted to working on Specific Aim 1. We submitted documents for ACURO approval (Major Task 1/Subtask 1) and approval was obtained. We completed all surgeries (castration and sham castration) and implanted placebo pellets and pellets containing leuprolide and flutamide (Major 1/Subtask 2). All mice in each experimental groups have been sacrificed for immunohistochemical studies and Western blots (Major Task 2/Subtask 1). Castration and the administration of flutamide and leuprolide are three methods used for androgen deprivation therapy in patients. We validated our experimental protocol by measuring plasma testosterone levels and the weight of the seminal vesicles in subjects in all treatment groups. We found that plasma testosterone levels and/or seminal vesicle weights were reduced significantly in the castrated, flutamide- and leuprolide-treated mice. We are in the process of completing the processing of the tissue (Major Task 2/Subtask 2) and data analysis (Major Task 2/Subtask 3). Because we have not completed the tissue processing and data analysis, we have no results to report at this time.

What opportunities for training and professional development has the project provided?

Nothing to report.

How were the results disseminated to communities of interest?

Nothing to report.

What do you plan to do during the next reporting period to accomplish the goals?

During the next reporting period we will complete the processing of the tissue (Major Task 2/Subtask 2) and data analysis (Major Task 2/Subtask 3). We also will begin Specific Aim 2, which will test the hypothesis that androgen deprivation disrupts cognitive behavior. We will follow the approved SOW and carry out surgeries and pellet implantation, initiate and complete behavioral testing and carry out analysis of the data. We expect to be able to submit a manuscript for publication by the end of the next reporting period.

4. IMPACT:

What was the impact on the development of the principal discipline(s) of the project?

Nothing to report.

What was the impact on other disciplines?

Nothing to report.

What was the impact on technology transfer?

Nothing to report.

What was the impact on society beyond science and technology?

Nothing to report.

5. CHANGES/PROBLEMS:

Changes in approach and reasons for change

Nothing to report.

Actual or anticipated problems or delays and actions or plans to resolve them

We encountered a problem that led to a delay in completing the processing of the tissue (Major Task 2/Subtask 2) and data analysis (Major Task 2/Subtask 3). All mice that are received from the vendor are subject to in-house quarantine for seven days. With one of our shipments several of the mice died within two days of arrival. We immediately contacted our resident veterinarian and he strongly suggested not to use the other animals in the shipment because they might be diseased and/or had undergone problems during shipping (e.g., over-heating and/or dehydration). A necropsy of the dead mice yielded no definitive findings. Subsequently we ordered another shipment of mice, but once again several mice died within two days of arrival and our veterinarian suggested that we not use the other mice in the shipment. At this point we contacted the veterinarian at the vendor, Charles River Laboratories, and he requested that we send them the dead mice for necropsy and histological analyses. After several weeks the reports came in and there was no definitive finding of disease or other reasons for the unexpected death of the mice. Although we considered switching to another vendor, this would cause problems because mice from different vendors can show subtle differences that could affect or confound the interpretation of the data. We have requested that the vendor ship fewer mice per shipping container and so far the mice have arrived in good shape. We do not anticipate having this problem with future orders, but it did cause a modest setback in our progress on Specific Aim 1 and currently we are working on catching up.

Changes that had a significant impact on expenditures

Nothing to report.

Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents
Nothing to report.

Significant changes in use of biohazards and/or select agents
Nothing to report.

6. PRODUCTS:

Publications, conference papers, and presentations
Nothing to report.

Journal publications.
Nothing to report.

Books or other non-periodical, one-time publications.
Nothing to report.

Other publications, conference papers and presentations.
Nothing to report.

Website(s) or other Internet site(s)
Nothing to report.

Technologies or techniques
Nothing to report.

Inventions, patent applications, and/or licenses
Nothing to report.

Other Products
Nothing to report.

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS:

What individuals have worked on the project?

Robert N. Pechnick, Ph.D. – no change

Tuerxun Ailikemu, M.D., Ph.D. – no change

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?
Nothing to report.

What other organizations were involved as partners?
Nothing to report.

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS:

N/A

QUAD CHARTS:

N/A

9. APPENDICES:

N/A